

**METALS AND TRACE ELEMENTS IN LIVERS OF
AMERICAN WHITE PELICANS
AT ANAHO ISLAND, NEVADA 2004**

By

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December 2004

Introduction

A large colony of American white pelicans (*Pelecanus erythrorhynchos*) nests on Anaho Island National Wildlife Refuge (NWR), which is on Pyramid Lake, Nevada, at the terminus of the Truckee River. Reproductive success of the colony was monitored in 1996 and a single egg was collected from each of 30 nests for analysis of organochlorine pesticides and metals and trace elements (Wiemeyer et al. 2001). Additional samples collected for residue analysis included: blood and feathers from two different age groups of nestling pelicans; livers from healthy and weak nestlings; liver and muscle samples from adults found dead or debilitated in the area; regurgitate samples from nestling pelicans; and fish from known feeding areas that were frequented by the pelicans. Reproductive success of the Anaho Island colony was normal based on hatching rates of eggs and survival of nestlings. Organochlorine and polychlorinated biphenyl (PCB) residues in eggs were below known effect levels, with biologically insignificant shell thinning. Organochlorine pesticides and PCBs were seldom detected in fish, but organochlorine pesticides were elevated in muscle samples of some adult pelicans. Mercury concentrations in eggs were generally below known effect concentrations, as were concentrations of other metals and trace elements. Metal and trace element concentrations in fish ranged widely for some constituents, with mercury of greatest concern. Microscopic lesions of mercury toxicity were absent in pre-fledging nestlings. Some adult pelicans had elevated mercury concentrations in their livers. Pre-fledging nestlings generally had much lower concentrations in their livers in both 1996 and in 1992 than those found in adults. The potential toxic effects in adults were difficult to evaluate because the proportion of methyl-mercury declined as the total mercury concentrations increased, thereby possibly providing protection from toxicity (Henny et al. 2002).

Pelican nesting success at Anaho Island has fluctuated greatly in the past eight years, from a high of 8,500 juveniles fledged in 1999 to a low of 440 juveniles fledged in 2003. Between 1996 and 1999, an average of 6,700 pelican nests produced 5,650 fledged juveniles, annually. Between 2000 and 2003, the averages dropped to only 4,200 pelican nests producing 630 juveniles each year. Drought conditions are thought to be a primary factor in high rates of juvenile mortality due to starvation. However, there have been lingering concerns regarding the potential adverse effects of mercury on reproduction and survival of American white pelicans nesting at Anaho Island. These concerns are greater in drought years when the birds may use more highly contaminated food sources. These concerns and the recent declines in the nesting population linked with reproductive problems warranted additional study of metals and trace elements, especially mercury, in eggs and livers of pre-fledging pelicans at the colony. The concerns in relation to mercury are heightened by an apparent increase in mercury concentrations in forage fish in Pyramid Lake, including cui-ui (*Chasmistes cujus*), tui chub (*Gila bicolor*), and Tahoe sucker (*Catostomus tahoensis*) between 1996 (Wiemeyer et al. 2001) and 2001 (Slotton and Ayers 2002). The cause of the increase is unknown, but might be related to increased mercury inputs into the lake in relation to the flood of January 1997 on the Truckee River.

The overall purpose of the study was to determine if inorganic contaminants may be responsible for the recent declines in reproductive success of American white pelicans that nest on Anaho Island. The objective of this portion of the study was to determine present concentrations of metals and trace elements in livers of pre-fledging nestling pelicans from Anaho Island.

Methods

Prefledging nestling pelicans that were moribund or that had recently died were collected by refuge personnel on July 28 and August 4 and 17, 2004 at Anaho Island NWR (Table 1). An adult pelican was also collected on April 9, 2004 by Dawn Graboski and Anna Keysers, Pyramid Lake Paiute Tribe. The birds were frozen shortly following collection. The birds were thawed on August 31 to September 2, 2004, the liver from each bird was removed with clean stainless steel instruments and placed in separate chemically-clean glass jars, and refrozen. The liver from the adult and a subset of the livers from nestlings, with representation from different dates of collection that appeared to have lesser degrees of postmortem decomposition, were submitted for analysis of metals and trace elements at Laboratory and Environmental Testing, Inc., Columbia Missouri on September 21, 2004.

Samples were homogenized at the analytical laboratory. Sub-samples were digested using various means, depending on the element to be analyzed. Arsenic and selenium analyses were by hydride generation atomic absorption (AA), mercury analyses were by cold vapor AA, lead analyses were by graphite furnace AA, and the remainder of the elements were analyzed by inductively coupled plasma (ICP). Quality assurance and quality control (QA/QC) included the analyses of one sample each of blank, duplicate, spike, and reference material. All QA/QC results were satisfactory and were certified by the Patuxent Analytical Control Facility of the U.S. Fish and Wildlife Service, Laurel, Maryland.

All residue concentrations were \log_{10} transformed prior to statistical analysis. One-half of the detection limit was assigned to samples with non-detectable concentrations. Geometric mean concentrations are reported only when $\geq 50\%$ of the samples had detectible concentrations of a given metal or trace element. SYSTAT[®] (SYSTAT 2000) programs were used in conducting analyses, which included data collected in prior years. One-way analysis of variance (ANOVA) was used for comparisons among years for nestlings only. Bonferroni multiple comparison tests were used to determine which years were significantly different from one another when differences ($p < 0.05$) among years were found with ANOVA.

All residue concentrations are reported in micrograms per gram ($\mu\text{g/g}$) on a dry weight basis, unless indicated otherwise.

Results and Discussion

General Condition

Information on the body weight, weight of liver sample taken for analysis, general condition (including postmortem condition), and a listing as to whether the liver was submitted for analysis for all birds collected in 2004 is provided in Table 1. Percent moisture for the liver from the adult (WPA0412) was 71.7. Percent moisture in the livers of the nestlings ranged from 73.5 to 76.9 with a mean of 75.8. All nestlings collected in 2004 were severely emaciated. Data on body weight was not collected for the nestling pelicans collected in 1992 or 1996. However, of the three nestlings that were collected in 1996 as representing normal young, two were rated as being in fair flesh, with the third rated as being in excellent flesh, whereas of the three debilitated birds collected in 1996, one each was rated as emaciated, in fair flesh, and in good flesh. All of the nestlings collected in 1992 were said to be emaciated. Collection dates in 1992 and 1996 were on August 28 and July 17, respectively. Although collection dates varied among years, the ages of the nestlings were assumed to be generally similar.

Comparison of Concentrations Among Years

Concentrations of metals and trace elements in livers of nestling pelicans from Anaho Island were compared among years, including previous data from 1992 and 1996 (Table 2). No significant differences among years were found for concentrations of arsenic, copper, molybdenum, and strontium. Concentrations of iron, magnesium, mercury, selenium, vanadium and zinc were significantly lower in 1996 than in either 1992 or 2004 with no significant differences between 1992 and 2004. Boron and chromium concentrations were significantly different between all years, with the concentrations in 1996 being the highest. Manganese concentrations were significantly different between 1992 and 1996, with the concentrations in 2004 not being different from either 1992 or 1996. Concentrations of aluminum, barium, beryllium, cadmium, nickel, and lead were detected in less than 50% of all samples, precluding statistical analyses.

Similar concentrations of many metals and trace elements between 1992 and 2004 may be due to several factors. First, both were drought years and adult pelicans may have been foraging in similar areas both years due to water conditions. Second, the birds collected in these years were in poor condition, being a mix of weak (euthanized) and recently dead birds in 1992 and recently dead or moribund (one) birds in 2004, with all being emaciated in both years. Wasting of muscle might result in the mobilization of metals and trace elements, resulting in increased concentrations in liver (see discussion on mercury below). This may be cause for concern in comparing data from 1992 and 2004 when the birds were emaciated, with that of 1996 when the condition of the nestlings was generally better. No significant differences in residue concentrations occurred between weak (euthanized) and recently dead nestlings in 1992 or between debilitated and healthy nestlings in 1996 (Wiemeyer et al. 2001). However, small sample sizes may have precluded the detection of significant differences. The lone emaciated nestling collected in 1996 had the highest or second highest concentration of 12 of 14 metals or trace elements that were detected that year.

The following concentrations ($\mu\text{g/g}$) were found in the liver of the adult pelican (i.e., WPA0412): aluminum 5.0, arsenic <0.20, barium <0.20, beryllium <0.10, boron <2.0, cadmium 1.0, chromium <0.50, copper 61.2, iron 2700, magnesium 766, manganese 9.7, mercury 150, molybdenum <2.0, nickel <0.50, lead <0.20, selenium 39, strontium 0.20, vanadium <0.50, and zinc 185. Residue concentrations in this bird have been added to the data set for adults collected from 1989 to 1996 that were previously reported (Wiemeyer et al. 2001), with the revised geometric means provided in Table 2.

Comparisons to Known Effect Concentrations

Concentrations of metals and trace elements were compared to known effect concentrations from various published sources. No data for interpretation were found for barium, beryllium, iron, magnesium, manganese, and strontium; therefore, they are not discussed further.

Arsenic is rapidly excreted from avian tissues (Eisler 2000b) and concentrations in all livers from this study were well below concentrations in livers associated with adverse effects such as reduced weight gain and diminished reproductive success (Camardese et al. 1990; Stanley et al. 1994). Arsenic concentrations were similar to those found in livers of juvenile and

adult American coots (*Fulica americana*) from the Humboldt Wildlife Management Area (WMA) in the late 1980's and mid to late 1990's (Wiemeyer et al. 2004).

Boron concentrations in pelican livers from this study were similar to or lower than concentrations in control mallards (*Anas platyrhynchos*) in experimental studies and were lower than concentrations associated with elevated exposures of boron that were related to adverse effects (Smith and Anders 1989; Hoffman et al. 1990). Boron concentrations in pelican livers were also generally slightly lower than those found in livers of juvenile and adult American coots from the Humboldt WMA in the late 1980's and mid to late 1990's (Wiemeyer et al. 2004).

Cadmium concentrations $>3 \mu\text{g/g}$ (dry weight) in liver were considered indicative of increased environmental exposure, whereas adverse effects were expected at concentrations $> 40 \mu\text{g/g}$ (Eisler 2000a). Only one adult pelican collected earlier (i.e., 1996) had a concentration (i.e., $3.1 \mu\text{g/g}$) barely exceeding the concentration associated with increased exposure.

Chromium concentrations in tissues of wildlife $> 4 \mu\text{g/g}$ were associated with probable exposure (Eisler 2000a). All chromium concentrations in pelican livers were below this level.

The no effect level of copper in bird liver is $< 60 \mu\text{g/g}$, with the toxicity threshold being $> 540 \mu\text{g/g}$ (U.S. Department of the Interior 1998). All copper concentrations in livers of nestling pelicans collected in 2004 exceeded $60 \mu\text{g/g}$; the concentration in the liver of the adult collected in 2004 only slightly exceeded this level. Copper concentrations in livers of nestling or immature ospreys (*Pandion haliaetus*) were often higher than those found in adults (Wiemeyer et al. 1980, 1987).

Mercury concentrations in bird livers between 1 to $10 \mu\text{g/g}$ wet weight (about 4 to $40 \mu\text{g/g}$ dry weight assuming 75% moisture content) were considered normal; however, concentrations of > 5 to $6 \mu\text{g/g}$ wet weight (about 20 to $24 \mu\text{g/g}$ dry weight) may be toxic to sensitive species, which would include mallards and other waterbirds (Heinz 1979; U.S. Department of the Interior 1998; Eisler 2000a). Heinz (1996) estimated that 20 to $60 \mu\text{g/g}$ (wet weight) mercury in livers was associated with harmful methylmercury exposure in adult birds; comparable dry weight concentrations would be approximately four times higher or about 80 to $240 \mu\text{g/g}$. The mercury concentration in the liver of only one nestling pelican in 2004 exceeded $80 \mu\text{g/g}$, which is cause for concern. The mercury concentration in the liver of the adult collected in 2004 also exceeded this level.

For comparative purposes, livers of nestling/fledgling double crested-cormorants (*Phalacrocorax auritus*) (another species in the Order Pelecaniformes) from Lahontan Reservoir (a mercury contaminated system) in 1998 had a geometric mean of $10.88 \mu\text{g/g}$ mercury (wet weight; about $43.5 \mu\text{g/g}$ dry weight) (Henny et al. 2002). The mean hepatic mercury concentration in this species from a reference area (Ruby Lake NWR) was $1.84 \mu\text{g/g}$ (wet weight; about $7.4 \mu\text{g/g}$ dry weight). Spleens from the birds from the contaminated area were significantly enlarged when compared with those from the reference area (means of 6.6 g vs. 2.6 g); this was consistent when evaluated in relation to total body weights. Spleen enlargement was significantly correlated with total mercury concentrations in liver. Henny et al. (2002) indicated that the spleen is an important site of methyl mercury demethylation in laboratory rodents. Young cormorants from the contaminated area had severe inflammation in the liver and lymphoid depletion in the thymus and bursa, with the reverse being found in the spleen. These birds also had smaller bursal follicles and severe vacuolar degeneration and inflammation of peripheral nerves. Mercury-related oxidative stress was also found in these birds, which included hepatic thiobarbituric acid reactive substances that are considered indicative of lipid

peroxidation and possible cellular damage. All of these findings may be of significance when examining the data from the nestling pelicans, especially when one considers the similar hepatic mercury concentrations in the two studies. However, the wasting of muscle of sick birds, as was present in the nestling pelicans, could result in the release of mercury from muscle and its further accumulation in liver (Scheuhammer et al. 1998). Therefore, our data must be interpreted with caution. The lack of data on body weights in 1992 and 1996 somewhat hinders a complete evaluation of the data. It would be helpful to collect samples of healthy nestling pelicans from Anaho Island in a drought year and an uncontaminated reference colony to study mercury residues in livers, organ weights, histopathology, and biochemistry.

Molybdenum concentrations in the liver of birds of 22-36 $\mu\text{g/g}$ have been associated with toxic effects (U.S. Department of the Interior 1998). Molybdenum concentrations in the livers of the nestling pelicans collected in 2004 were approximately an order of magnitude lower, with an even lower concentration in the liver of the adult collected in 2004.

Adverse effects to birds were associated with nickel concentrations in liver $> 3 \mu\text{g/g}$ (Eisler 2000a). Nickel was not detected in any of the livers of pelicans collected in 2004.

Lead concentrations in livers of waterfowl $> 6 \mu\text{g/g}$ were considered elevated (Eisler 2000a). Lead was not detected in any livers of pelicans collected in 2004.

The median concentration of selenium in livers of birds classified as carnivores was 8.2 $\mu\text{g/g}$ (U.S. Department of the Interior 1998), with concentrations < 5.2 to $< 10 \mu\text{g/g}$ classified as being acceptable and concentrations $> 10 \mu\text{g/g}$ being associated with adverse effects (Eisler 2000b). Selenium concentrations in three of the nine nestling pelicans collected in 2004 had $> 10 \mu\text{g/g}$ selenium, with the highest concentration being 31 $\mu\text{g/g}$.

Information on the interpretation of vanadium concentrations in livers of birds is very limited. The lipid metabolism of mallard hens fed a diet containing 100 $\mu\text{g/g}$ of vanadium was altered (White and Dieter 1978). The birds on this dietary concentration had a mean of 0.657 $\mu\text{g/g}$ vanadium (wet weight) in livers, with only 0.019 $\mu\text{g/g}$ (wet weight) in livers of controls. The higher level was considered a level of concern. Vanadium concentrations of nestling pelicans exceeded this level.

Liver concentrations of zinc in various species of birds of 21 to 33 $\mu\text{g/g}$ were considered normal, whereas livers of zinc poisoned birds had 75 to 156 $\mu\text{g/g}$ (Eisler 2000a). However, liver concentrations of $< 210 \mu\text{g/g}$ were considered to be of no effect by the U.S. Department of the Interior (1998), with concentrations $> 2,100$ being the toxicity threshold. Zinc concentrations in all livers from nestling pelicans collected in 2004 exceeded 210 $\mu\text{g/g}$; however, none exceeded 2,100 $\mu\text{g/g}$.

Feeding Areas

Aerial surveys for waterfowl, which included counts of white pelicans, were conducted by the Nevada Department of Wildlife (N. Saake, Nevada Department of Wildlife, pers. comm.) on May 28 to June 2 and on July 23, 2004. Data from these counts are compared to those conducted during the same months in 1996 (Wiemeyer et al. 2001; Table 3). Concentrations of metals and trace elements in livers of nestling white pelicans at Anaho Island may vary from year to year in relation to where the parent birds are feeding. However, it is unknown if the birds that were counted were parents feeding young at the nesting colony in either year. Therefore, caution should be used in interpretation of the data in relation to liver residues in the nestlings. The overall percentage of pelicans found at Walker Lake in mid- to late-July was much higher in

2004 than in 1996, with the reverse being true at Pyramid Lake. This may be related to the poor reproductive success in 2004 involving the abandonment of many nesting attempts.

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Table 1. American White Pelicans collected in 2004 for possible metal and trace element analysis of livers.

Number	Collection Location	Date	Age ^a	Sex ^b	Body Weight (g)	Liver Weight (g)	Comments	Analyzed
WPJ0401	Anaho Island, A Colony shoreline	8/04/04	Nr. Juv.	Unk.	2628	37.7	Emaciated; poor postmortem condition	No
WPJ0402	Anaho Island, A Colony shoreline	8/04/04	Nr. Juv.	Unk.	1785	39.8	Emaciated; fair to good postmortem condition	Yes
WPJ0403	Anaho Island, A Colony shoreline	8/04/04	Nr. Juv.	Unk.	2705	52.9	Emaciated; good postmortem condition	Yes
WPJ0404	Anaho Island, A Colony shoreline	8/04/04	Nr. Juv.	Unk.	2630	42.7	Emaciated; fair to good postmortem condition	No
WPJ0405	Anaho Island, A Colony shoreline	7/28/04	Nr. Juv.	Unk.	2606	49.2	Emaciated; good postmortem condition	Yes
WPJ0406	Anaho Island	8/17/04	Nr. Juv.	Unk.	2759	30.0	Emaciated; fair to good postmortem condition	No
WPJ0407	Anaho Island	8/17/04	Nr. Juv.	Unk.	2640	28.3	Emaciated; fair postmortem condition	No
WPJ0408	Anaho Island	8/17/04	Nr. Juv.	Unk.	1887	36.2	Emaciated; fair to poor postmortem condition	No
WPJ0409	Anaho Island	8/17/04	Nr. Juv.	Unk.	2822	36.6	Emaciated; fair postmortem condition	No
WPJ0410	Anaho Island	8/17/04	Nr. Juv.	Unk.	2533	45.6	Emaciated; fair to good postmortem condition	Yes

Table 1. American White Pelicans collected in 2004 for possible metal and trace element analysis of livers. (continued)

Number	Collection Location	Date	Age ^a	Sex ^b	Body Weight (g)	Liver Weight (g)	Comments	Analyzed
WPJ0411	Anaho Island	8/17/04	Nr. Juv.	Unk.	2192	37.8	Emaciated; good postmortem condition	Yes
WPA0412	Near Nixon, Nevada	4/09/04	Adult	Unk.	>6800 ^c	75.1	Moderate subcutaneous fat and heavy abdominal fat. Excellent postmortem condition. Cause of death unknown; no evidence of trauma found.	Yes
WPJ0413	Anaho Island	8/17/04	Nr. Juv.	Unk.	2578	45.0	Emaciated; good postmortem condition.	Yes
WPJ0414	Anaho Island	8/17/04	Nr. Juv.	Unk.	2510	36.8	Emaciated; fair to good postmortem condition	Yes
WPJ0415 ^d	Anaho Island	8/17/04	Nr. Juv.	Unk.	2735	47.6	Emaciated; good postmortem condition. Believed to have been collected alive.	Yes
WPJ0416 ^d	Anaho Island	8/17/04	Nr. Juv.	Unk.	2732	50.9	Emaciated; fair to good postmortem condition	Yes

^a Nr. Juv. = near juvenile; not believed to have fledged or be self sufficient.

^b Sex was not determined in any case with certainty.

^c The weight of WPA0412 could not be determined as it went above the capacity of the scale.

^d WPJ0415 and WPJ0416 were in the same bag with two different labels on the outside. WPJ0415 was assumed to have been the one collected alive due to its better post-mortem condition.

Table 2. Concentrations ($\mu\text{g/g}$ dry weight) of metals and trace elements in livers of pre-fledging nestling white pelicans collected from Anaho Island in 1992, 1996, and 2004, and adult white pelicans collected in northwestern Nevada in 1989-2004.

Element	Nestlings – Anaho Island			Adults
	1992 (n = 7)	1996 (n = 6)	2004 (n = 9)	1989-2004 (n = 12)
Aluminum	nd ^a (0) ^b (<3.6 - <4.7) ^c	nd (0) (<3.2 - <4.4)	3.0 (7) (<3.0 - 5.0)	2.9 (6) (<2.5 - 8.5)
Arsenic	nd (3) A ^d (<0.19 - 0.44)	0.33 (6) A (0.23 - 0.52)	0.30 (6) A (<0.20 - 1.4)	nd (5) (<0.13 - 0.60)
Barium	nd (0) (<0.72 - <0.95)	nd (0) (<0.64 - <0.88)	0.46 (8) (<0.20 - 1.2)	nd (0) (<0.20 - <0.68)
Beryllium	nd (0) (<0.07 - <0.09)	nd (0) (<0.06 - <0.09)	nd (0) (<0.10)	nd (0) (<0.05 - <0.10)
Boron	1.6 (5) B (<1.6 - 2.6)	3.5 (6) C (2.1 - 6.5)	nd (0) A (<2.0)	2.2 (9) (<2.0 - 4.2)
Cadmium	nd (2) (<0.22 - 0.46)	nd (1) (<0.19 - 0.20)	nd (4) (<0.10 - 0.30)	1.2 (12) (0.54 - 3.1)
Chromium	0.58 (7) B (0.56 - 0.61)	1.3 (6) C (1.0 - 1.9)	nd (0) A (<0.50)	0.94 (10) (<0.49 - 2.3)
Copper	105. (7) A (61 - 179)	83. (6) A (19 - 180)	159. (9) A (77 - 446)	37. (12) (14 - 74)
Iron	6571. (7) B (5562 - 8911)	1469. (6) A (881 - 2422)	6634. (9) B (3880 - 8660)	3498. (12) (2069 - 6284)
Magnesium	751. (7) B (663 - 822)	667. (6) A (611 - 737)	770. (9) B (635 - 858)	637. (12) (486 - 886)
Manganese	8.8 (7) A (7.2 - 11)	12. (6) B (8.3 - 15)	9.6 (9) AB (8.1 - 12)	9.0 (12) (5.7 - 11.4)

Table 2. (continued)

Element	Nestlings – Anaho Island			Adults
	1992 (n = 7)	1996 (n = 6)	2004 (n = 9)	1989-2004 (n = 12)
Mercury	34. (7) B (25-56)	10. (6) A (6.2-14)	39. (9) B (18-160)	60. (12) (12-461)
Molybdenum	3.1 (7) A (2.1-3.6)	2.7 (6) A (2.2-3.7)	2.9 (9) A (2.0-4.0)	1.9 (11) (<2.0-3.5)
Nickel	nd (1) (<0.43-8.0)	0.77 (6) (0.46-1.5)	nd (0) (<0.50)	0.65 (9) (<0.45-1.6)
Lead	nd (0) (<1.8-<2.4)	nd (0) (<1.6-<2.2)	nd (0) (<0.20)	nd (4) (<0.2-2.0)
Selenium	8.3 (7) B (5.2-12)	4.5 (6) A (3.8-5.4)	11. (9) B (5.9-31)	23. (12) (9.9-140)
Strontium	1.1 (7) A (0.46-2.0)	1.4 (6) A (0.52-2.4)	2.2 (9) A (0.68-5.1)	0.28 (10) (<0.20-0.53)
Vanadium	2.4 (7) B (1.7-3.0)	0.72 (6) A (0.49-1.4)	2.6 (9) B (1.0-5.3)	0.33 (7) (<0.45-0.65)
Zinc	665. (7) B (560-786)	257. (6) A (145-574)	750. (9) B (351-1230)	173. (12) (112-386)

^a Geometric mean; nd = not computed; <50% with detectable residues.

^b Number with detectable residues.

^c Extremes of residues.

^d Years (nestlings only) for a given element not having a capital letter in common were significantly different ($P < 0.05$)

Table 3. Distribution of American white pelicans on western Nevada lakes and wetlands during May-July aerial survey dates in 1996 and 2004. Data provided by Norm Saake of the Nevada Department of Wildlife.

Locations	Aerial survey dates				
	1996			2004	
	16-24 May	11 June	16 July	28 May-2 June	23 July
<u>Truckee River Basin</u>					
Pyramid Lake	9,668 (73) ^a	6,260 (60)	2,633 (33)	3,940 (75)	174 (9)
Washoe Lake	3	22	40	265 (5)	150 (8)
<i>Subtotals</i>	<i>9,671 (73)</i>	<i>6,282 (60)</i>	<i>2,673 (34)</i>	<i>4,205 (80)</i>	<i>324 (17)</i>
<u>Carson River Basin</u>					
Lahontan Reservoir	238 (2)	127 (1)	316 (4)	228 (4)	82 (4)
Carson Lake	1,072 (8)	847 (8)	1,043 (13)	89 (2)	33 (2)
Stillwater NWR	307 (2)	190 (2)	1,875 (24)	-	-
Sheckler Reservoir	116	36	56	-	-
S-Line Reservoir	6	48	61	-	0
Harmon Reservoir	-	8	52	15	16
Canvasback Gun Club	110	96	288 (4)	19	145 (8)
Indian Lakes	310 (2)	4	44	-	-
Leter Reservoir	80	1	183 (2)	2	97 (5)
Other areas	5	-	109 (1)	80 ^b (2)	112 ^c (6)
<i>Subtotals</i>	<i>2,244 (17)</i>	<i>1,357 (13)</i>	<i>4,027 (51)</i>	<i>433 (8)</i>	<i>485 (25)</i>
<u>Walker River Basin</u>					
Walker Lake	1,131 (9)	2,779 (26)	761 (10)	435 (8)	890 (46)
Mason Valley WMA	73	23	143 (2)	25	94 (5)
Weber Reservoir	4	6	25	19	30 (2)
Alkali Lake WMA	6	-	-	-	2
Topaz Lake	52	-	-	63 (1)	34 (2)
Other areas	-	-	-	1	5
<i>Subtotals</i>	<i>1,266 (10)</i>	<i>2,808 (27)</i>	<i>929 (12)</i>	<i>543 (10)</i>	<i>1,055 (55)</i>

Table 3. (continued)

Locations	Aerial survey dates				
	1996			2004	
	16-24 May	11 June	16 July	28 May-2 June	23 July
<u>Lower Humboldt River Basin</u>					
Humboldt WMA	-	0	198	-	0
Rye Patch Reservoir	-	33	33	31	57 (3)
Humboldt River	-	40	8	28	-
<i>Subtotals</i>	-	73 (<1)	239 (3)	59 (1)	57 (3)
<i>Total Observed</i>	13,181	10,517	7,868	5,240	1,921

^a Percent of total counted for a given date provided in parentheses.

^b On Stillwater Wildlife Management Area.

^c 108 on Stillwater Wildlife Management Area, 3 on Indian Lakes, 1 in Carson Valley.